Then it is highly probable that pinealocytes exposed to modulated RF/MW will exprience an outflow of calcium ions, a reduction of the cAMP signal and a reduction in the production of melatonin. This is a highly plausible mechanism to explain why RF/MW can reduce pineal melatonin production with all the adverse health consequences which flow from that.

3.2 The Max Planck Institute Reports Circadian Rhythm Effects:

Human biometeorology has a great deal to teach us about the effects of natural electromagnetic fields and biological reactions. There is a strong evidence, Wever (1974), that natural ELF signals such as the Schumann Oscillations, coupled with the earth's magnetic field, help to phase lock the 24 h circadian rhythm in people and animals. Isolation experiments show that the dark/light cycle is insufficient to fully regulate the circadian rhythm but other environmental stimuli, called "Zeitgebers" by the German researchers, are also required to synchronize the rhythm. These must be globally available, naturally occurring signals since almost all terrestrial life is tuned to the 24 hr cycle.

The day/night light cycle is the primary driver of circadian rhythm. for when people are deprived of this light cycle their daylength drifts, generally becoming longer. An extensive research programme has been carried out by the Max Planck Institute over several years and involving over 200 subjects.

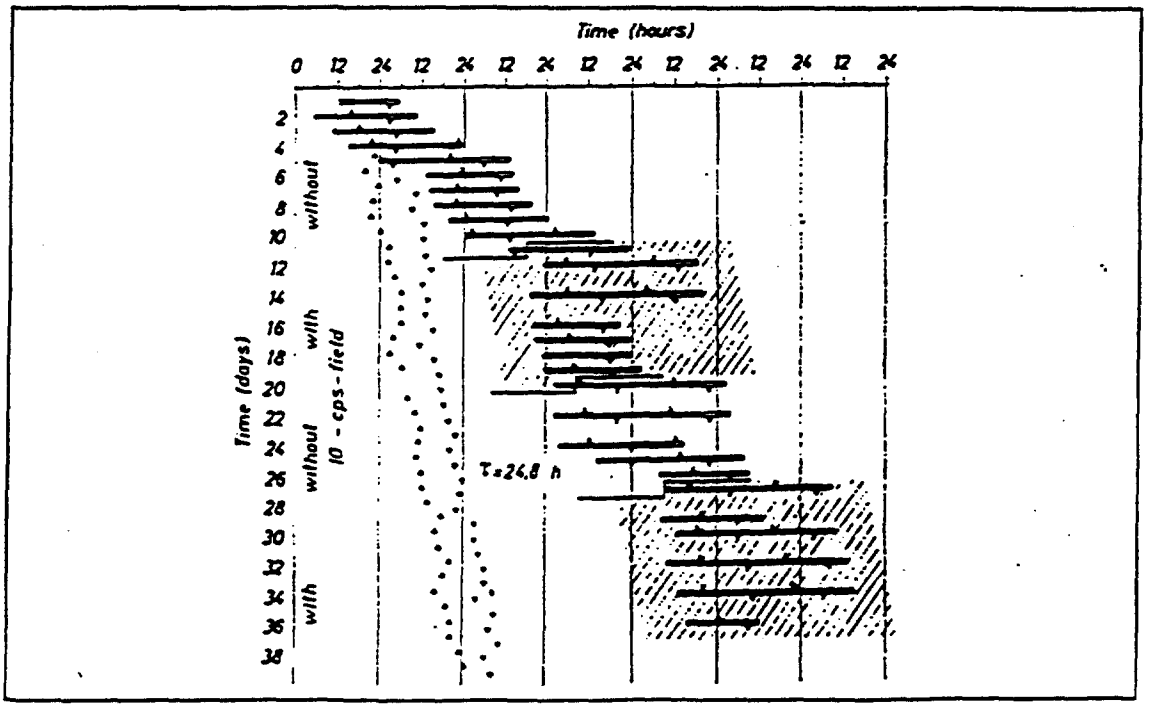

Figure 8: Free-running circadian rhythm of a subject living under strict isolation from environmental time cues, during the first and third section protected from natural and artificial electromagnetic fields, during the second and fourth sections under the influence of a continuously operating 10 Hz electric field of 2.5 V/m, Wever (1970).

Two isolation rooms were used, one of which was also shielded from environmental electromagnetic fields (Room 2). In a simple "free running" experiment, it was found that the mean day period was 24.87 h for Room 1 and 25.26 h for Room 2. The difference between the light isolated (Room 1) and the light and electromagnetic field isolated subjects (Room 2) was significant at the $p<0.01$ level, Figure 8. Those isolated from the extremely small environmental electromagnetic fields had mean daylengths that were significantly longer and more variable. The standard deviation of their variation in daylength was also significantly larger Room 2 and the number of internal desynchonizations was greater in Room 2 with $p<0.001$.

Depriving people of access to the natural electromagnetic fields made a very significant difference in their daily rhythm. Other experiments with a very low level artificial ELF signal were carried out. This 2.5 V/m (peak-to-peak) 10 Hz signal (rms-amplitude of 1.77 V/m giving $S=0.83\mu W/cm^2$) reduced the desynchronization significantly ($p<0.001$). In many experiments, no case of internal desynchronization occurred as long as the 10 Hz field was in operation, Weber (1969). Static electric fields were investigated but showed no effects.

Weber (1974) concludes that their research gives:

> **"significant proof that electromagnetic fields in the ELF range influence the human circadian rhythms and therefore human beings."**

People who were deprived of the light/dark cycle and natural electromagnetic fields with intensities of the order of 0.3 pW/cm^2 showed significant shifts in circadian rhythm while an artificial ELF field of $0.8\mu W/cm^2$ significantly reduced the desynchronization, mean period and variance of the circadian rhythm.

The biological mechanism involved in brain detection of extremely low intensity ELF signals is not discussed by Wever. This substantial project, carried out by a prestigious laboratory, establishes that human beings have the ability to sense and react to extremely small electromagnetic signals.

3.3 Biological Sensors or Environmental Fields:

Biological systems are sensitive to external EM fields for many functions. There is unequivocal experimental evidence that fields from ELF to UHF (10 Hz to 450 MHz) interact directly with brain tissue, Adey (1981). Dr Adey cites bird navigation, bird circadian rhythms, monkeys' subjective time estimations and human circadian rhythms which are all related to tissue level gradients of about 10^{-7} V/cm. RF/MW radiation interacts with brain tissue by causing single- and double-strand DNA breakage, Lai and Singh (1995, 1996) and by changing the human EEG, Von Klitzing (1995).

The fact that intrinsic cell neuroelectric gradients are far higher than these observed tissue gradients, e.g. Membrane Potential 10^5 V/cm, Synaptic Potential 10^3 V/cm and Electro-encephalogram 0.02 to 0.05 V/cm, attests to the vital role of modulation of EMR and the existence of amplification processes at the cellular level, Adey (1989).

Induction of electric fields in tissue at the cellular level varies with the intensity and the nature of the environmental field. Typical endogenous EM fields, with ELF modulation,

induce fields in the order of 10^{-1} to 10^{-7} V/cm in the pericellular fluid (fluid surrounding the cell). RF/MW fields penetrate the organ or body much more effectively than the ELF fields. For example, when chick brains were exposed to an applied 56 V/m ELF field 1-32 Hz, they induced a tissue gradient of 10^{-7} V/cm. However, when a 56 V/m RF field (147 MHz, 0.8 mW/cm^2, ELF modulated) was applied, it produced a tissue gradient of 10^{-1} V/cm., Bawin and Adey (1976). Both of these signals significantly changed the calcium ion efflux from the chick brain tissue.

Thus the RF/MW field produced a cellular tissue gradient 1 million times higher than the ELF field of the same external field strength. This shows the highly penetrative nature of RF/MW fields compared to ELF fields. Since the energy flux relates to the square of the electric field gradient strength, Eq. 3, the energy imparted to the cell tissue by RF/MW modulated radiation as many orders of magnitude higher than the same external field strength of ELF EMR.

4. Cellular Biology:

4.1 Introduction:

The fundamental basis of biologic activity is the cell. Cell biology and biochemistry has advanced our understanding of cell behaviour and cellular processes to highly advanced levels. The structure of cells is well described, the processes which regulate cell growth and development, the genetic basis of cell reproduction and the amino acid typing of complex molecules, including RNA and DNA is being advanced daily. This shows the importance of understanding and appreciating cellular characteristics and processes in order to understand the interactions of EMR with living tissue and potential health hazards from EMR exposure.

In cellular aggregates that form tissues of higher animals, cells are separated by narrow fluid channels that take on a special importance in signaling from cell to cell. These channels act as windows on the electrochemical world surrounding each cell. Hormones, antibodies, neurotransmitters and chemical cancer promoters, for example, move along then to reach binding cells on the cell membrane receptors.

Molecular and cell level electric charges, and electric and magnetic fields play a fundamental role in these processes and in the organisation of the complex cellular and macrocellular structures. With these understandings has come new insights. Gone from the outset are notions of isotropy. We see schemes of biochemical and biophysical organisation of unparalleled complexity. Concepts of linear systems disappear and are replaced by non-linear, non-equilibrium thermodynamics. Observations and models are consistent with quantum processes involving long-range interactions between electrical charges on cell surface macromolecules, Adey (1992c).

It is at the level of the cell that non-thermal effects become very evident.

It has been asserted by some that thermal noise, expressed in the term kT as a function of the Bolzmann constant and the absolute temperature, must remain a monolithic threshold below which no biological threshold can exist. Some biophysicists and others still hold this view despite the wealth of physiological evidence that sensory thresholds

descend substantially below the floor of thermal noise, as happens for example in the auditory system of the ear, Adey (1993). Also, attention is now directed to newly defined roles for free radicals, that may also participate in highly cooperative detection of weak electromagnetic fields, "even at levels below the thermal (kT) noise, McLaughlan (1992), and Grundler et al. (1992).

4.2 Cell-based resonant absorption:

Water is known to strongly absorb microwaves. This characteristic is used in microwave cooking. Living biological tissue is water-rich. It is reasonable then to expect that water in tissue will absorb microwave energy. The surprise is the response to absorbed RF radiation. During the MacIntyre Case, the Court was not told about the research of Liu and Cleary (1995) in which they showed, using classical scattering theory, that both radiofrequency (RF) and microwave (MW) radiation is resonantly absorbed in the bound water layers on the cell membrane.

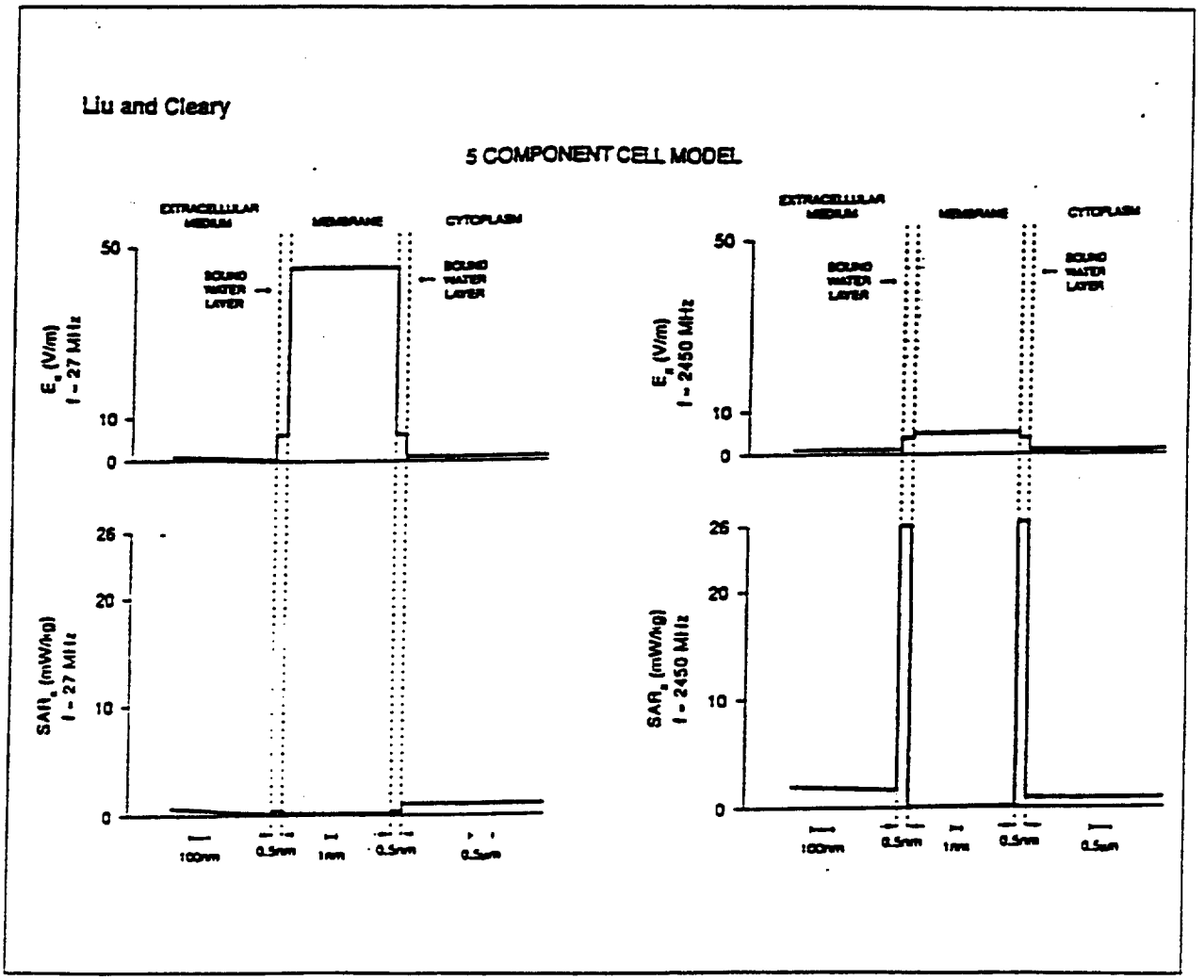

Figure 9: Spatial distribution of the maximum induced E-filed component(E_x) and SAR (SAR_x) on the x-axis of a five component mammalian cell model (extracellular medium, bound-water layer on exterior cell membrane surface, cell membrane, bound-water layer on inside surface of cell membrane surface, cytoplasm) exposed to 27 or 2450 MHz continuous plane-wave electromagnetic radiation. Incident field strength, 1 V/m. ($0.3\mu W/cm^2$)

This creates an electric field across the cell membrane in the case of RF and thermal fields in the case of MW, Figure 9.

This research provides vital link between the whole body-absorption of RF/MW radiation by human beings and animals and the altered cellular biochemistry demonstrated in isolated cell lines in laboratory experiments. It is another direct rebuttal of the thermal view because the effect varies strongly with frequency between electric field and heating.

Liu and Cleary (1995) have shown, in a biophysical model of cells, that the bound water layer on each side of the cell membrane, resonantly absorbs radiofrequency and microwave radiation at the surface of the cell membrane.

There is a vast difference between the effect of an RF (27 MHz) signal and a microwave (2.45 GHz) signal. For the 27 MHz signal the cell has a massive change in the electric field in the membrane and very little heat absorption. For the 2.45 GHz microwave signal there is a very small change in the electric field from the outside to the inside of the cell, but there is a massive, resonant absorption of heat energy on the inside and outside surface of the cell due to the presence of the bound water layers.

I have asked Professors Liu and Cleary to re-run the model for 915 MHz, a common mobile phone frequency. The biophysics suggests that this would show a lower heating response compared to 2450 MHz and a lower electric field difference than the 27 MHz signal.

This very significant paper shows that classical physics predicts resonance absorption of RF/MW radiation at the cell level. Hence there are energy, thermal and electric gradients which can alter the biochemical and chemical reactions at the cellular level. Professor Cleary pointed out to me that this paper also shows a difference in the resonant absorption on the various axes of the cell, depending on the polarization of the incident EM radiation. In a spherical model these differences are as great as a factor of 13 above average. That means, he explained, a mean SAR of 0.007 W/kg could well have localized cellular based SARs or 0.10 W/kg. He was worried about the effects of cell phones on brain cells in the head near the antenna, in the light of this and his other work on cell-cycle timing changes and DNA changes in Chinese Hamster Ovary (CHO) cells, for example.
The absorption of the RF/MW energy at the cell membrane gives a basis to move on to the biochemistry of altered cellular processes, including the effect on signal transduction, cell cycle timing, on ODC in tumour development, on calcium ions and the immune system, and on melatonin in relation to free radical control and elimination. These in turn relate to adverse health effects such as cancer and immuno-competence, sleep disturbance, memory disfunction and concentration disruption.

4.3 Biochemistry and cell biology:

During the MacIntyre Case, Associate Professor Richard Luben presented evidence of biochemical mechanisms which are observed to change under exposure to modulate RF fields, Luben (1995). This included changes in calcium ion efflux and Ornithine Decarboxylase (ODC), both of which are involved in the signal transduction aspects of control of the growth and development of cells in the human body and other animals.

Two of Dr Luben's key statements are worth recalling. He stated that laboratory studies had shown the similarities and parallels in the biological effects of ELF and RF modulated by ELFs. For example, calcium ion efflux and ODC are observed to vary in similar ways in ELF fields and in RF fields modulated at ELF frequencies, Byus (1994), Giuliana, et al. (1996).

Dr Luben also stated that the electromagnetic radiation did not need to enter the cell in order to change its behaviour, it just needs to be absorbed by the cell surface, and then the altered signal transduction process changes the cell behaviour.

A more detailed and updated review of biophysics and biochemistry reveals clear means of EMR altering the biochemical behaviour at the cellular level with a great deal of detailed existing understanding about these processes. These form a set of plausible mechanisms to explain the way in which EMR can change cell behaviour and hence can cause adverse health effects. Where these are matched by appropriate epidemiology an extremely strong association is established. Where there is a pattern of epidemiology which is consistent with animal experiments and for which there are detailed biophysical and biochemical mechanisms, the evidence approaches the level required to establish cause and effect.

The research here reviews our current understanding of cell structure and processes, including resonant absorption of radiofrequency and microwave radiation at the cell membrane, gap junction communication between cells, signal transduction processes from regulating cell growth and behaviour, the vital role of calcium ions, the implications for changes in ODC, the formation and effects of free radicals, the dendridic structure of the brain which relates to neurotransmitters and EEG, and the role of neurotransmitters such as serotonin and adrenaline, and neurohormones such as melatonin.

Evidence will then be presented showing cellular and molecular changes which occur with exposure to electromagnetic radiation which are directly or indirectly related to known biophysical biochemical characteristics of cells. The implications of this for public health will be discussed. The identified biophysical and biochemical mechanisms will then be related to epidemiological research and public health implications will be discussed and exposure standards will be recommended where supportable by sound research.

4.4 Cell Biochemistry and Neurophysiology

The last 10 to 15 years has seen an exciting and challenging revelation of the complex biochemistry at the cellular and molecular level in living systems. As laboratory techniques have advanced we have progressed from the study of organs, to tissues, to cells and to molecules. The behaviour and organisation of cells and molecules in living tissue relies on sequences of reactions which use and transform energy in the continual creation and re-creation of molecular complexes including material to form cell walls (membrane), cell nuclei, inside to outside cell communications, cell to cell communications, enzymes to stimulate and slow cell division and cell growth, and the RNA and DNA molecules which code the genetic structure of the host of different types of cell, and cell and tissue structures which make up highly organized living systems.

4.5 The Cell:

The cell is an identifiable entity of all living organisms and is recognised as the fundamental unit of biologic activity. Cells consist of a nucleus which is surrounded by cytoplasm, which contains various organelles, and is enclosed in a cell or plasma membrane, Figure 10.

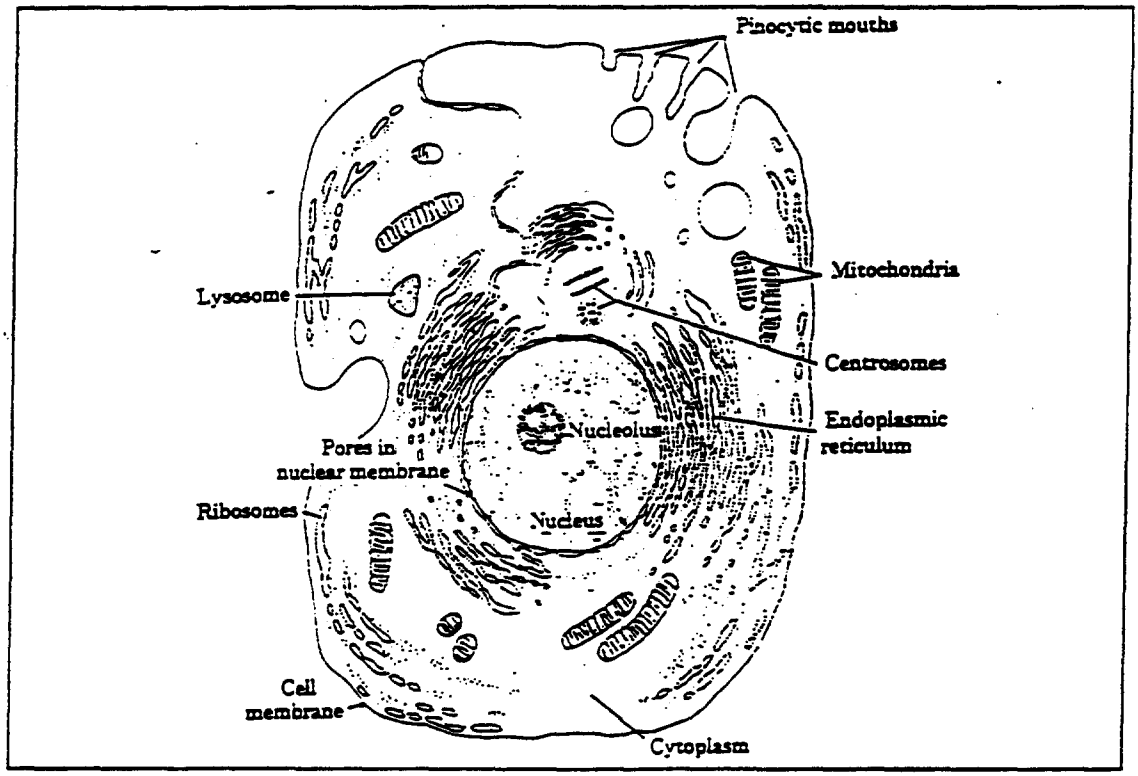

Figure 10: A typical animal cell (Dorland 28, p285)

The nucleus contains the hereditary material of DNA and chromosomes, along with the proteins and enzymes which are necessary for the sustenance of the nucleus and the processes of chromosome separation during mitosis.

The cytoplasm is the protoplasm of a cell exclusive of that nucleus. It consists of a continuous aqueous solution (cytosol) and the organelles and inclusions suspended in it. It is the site of most biochemical activities of the cell.

The cell membrane is a bimolecular layer of lipids which encloses the cytoplasm and nucleus. It had is permeable to some substances and contains protein structures which pass through the membrane, providing for processes such as signal transduction, see Figure 5 below.

Cells form many shapes and have a host of different functions. Some cells are bound together to form tissue such as in skin and muscles, some are near spherical and float in fluid, such as T-cells, and others are dendridic, with long dendrite structures extending to several times the diameter of the central cell body, such as many brain and central nervous system cells.

4.6 The Cell cycle:

A cell is a cooperative of molecules which is capable of reproducing itself. Cells are discrete entities that grow and divide. Most cells must complete four tasks during the cell cycle. They must grow, replicate their DNA, segregate their chromosomes into two identical sets and divide. To do this a cell needs between 2000 and 5000 different enzymes and structural proteins.

Some of the molecules, like ribosomal proteins and RNAs are present in the millions per cell, while DNA are present as only one or two. Cells contain many different types of proteins, each specialized for a particular role in the life of the cell. Important classes include enzymes that produce the building blocks for the synthesis of DNA, RNA and proteins, and the enzymes which build these blocks to replicate DNA, transcribe DNA into RNA, and translate mRNA into protein. The form and function of cells depend on the structural proteins that form the cytoskeleton and on the motor proteins that move objects along elements of the cytoskeleton, such as chromosomes. Mammals are estimated to have as many as 200 different cell types.

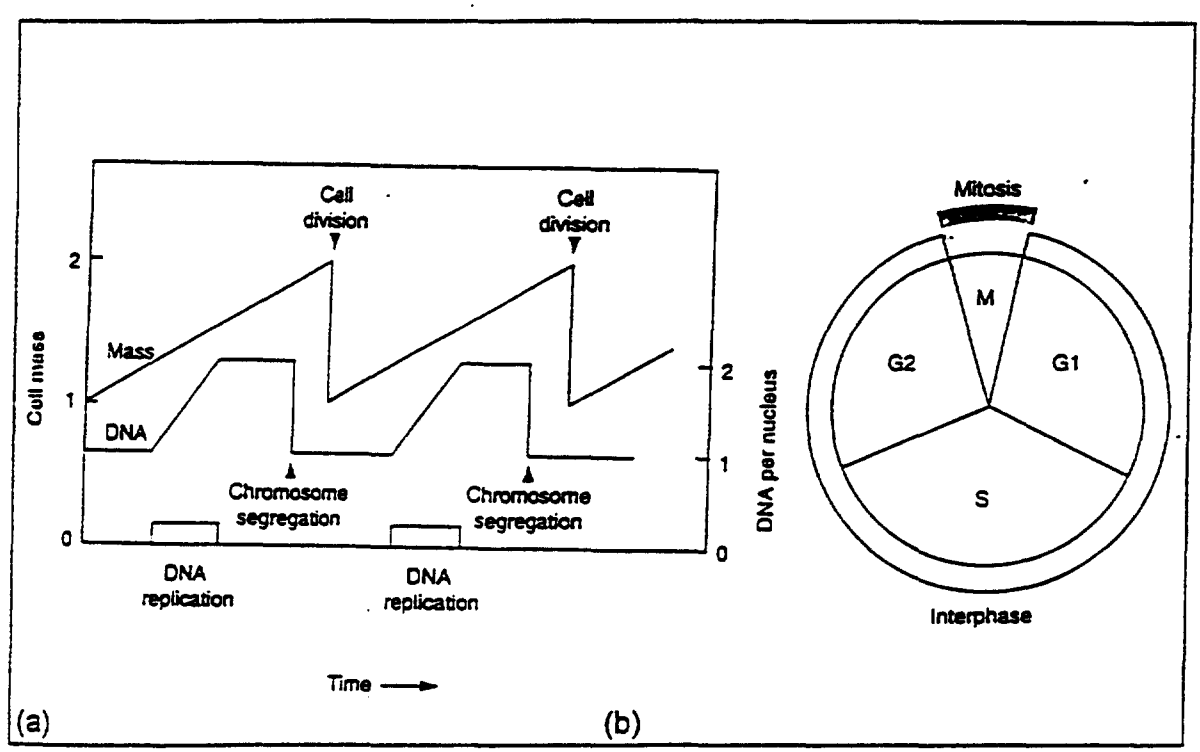

Figure 11: (a) Cell growth and DNA content during the cell cycle. Mass increases continuously throughout the cell cycle while DNA content is constant for most of the cycle, increasing during the S phase as DNA replicates, then falling dramatically during chromosome segregation.

(b) Stages of the cell cycle, which in adult vertebrates rapid cell cycles take about 24 hours. Mitosis (M) represents about 5 % of the cycle, and there is a substantial gap (G1) between mitosis and DNA synthesis (S), as well as a gap (G2) between replication and mitosis.

Source: The Cell Cycle, by Murray and Hunt (1993).

The cell cycle is divided into four discrete phases. Rapidly dividing human cells have a cell cycle which lasts about 24 hours.

The cell cycle is divided into two fundamental parts: interphase, which occupies the majority of the cell cycle, and mitosis, which lasts about 30 minutes, ending with the division of the cell. During interphase DNA is diffusely distributed throughout the nucleus, and individual chromosomes cannot be distinguished. Little activity can be seen in the microscope but two important classes of process are occurring, continuous processes (referred to collectively as 'growth') and stepwise processes which occur once per cycle.

For example, chromosome replication is restricted to a specific part of interphase called S phase (for DNA synthesis). S phase occurs in the middle of interphase, preceded by a gap called G1 and followed by a gap called G2, Figures 11a and 11b. After each chromosome has been replicated, the two daughter chromosomes remain attached to each other at multiple points along their length and are referred to as sister chromatids.

In a typical animal cell cycle, G1 lasts 12 hours, S phase 6 hours, G2 6 hours and mitosis (M) about 30 minutes.

The interaction mechanisms of radiation and some chemicals which have strong adverse health effects occur through alteration or interference with the cell cycle. X-ray radiation is known to damage cells and their DNA, while caffeine is known to accelerate mitosis. Rowley (1990) reported on studies into the repair of radiation-induced chromatid aberrations: relationship to G2 arrest in CHO cells. The literature suggests that the function of radiation-induced G2 arrest is to allow repair of potentially lethal damage before cell-entry into, and damage expression in, mitosis. The nature of the damage repaired is not known, but chromosome aberrations have been considered.

To examine this possibility in G2 cells, Rowley (1990) compared the rate of repair of chromatid aberrations in CHO cells progressing to or arrested in G2, with the rate of repair of the damage which gives rise to G2 arrest. To measure aberration repair rates, exponentially growing CHO cells arrested in G2 with 1.5, 2.5 or 3.5 Gy of X-rays were released into mitosis by treatment with 5 mM caffeine immediately or 1, 2 or 3 h after irradiation. Aberration frequencies in these cells were then related to the caffeine-free (repair) interval. To measure the rate of repair of arrest-causing damage a split-dose procedure was used. The half-times for aberration repair were approximately 1 h for achromatic gaps and 1.5 h for breaks, intrachanges and interchanges. The half-time for arrest damage repair varied with radiation dose. This result suggests that chromatid aberrations are not a primary cause of radiation-induced G2 arrest.

While Rowley (1990) has shown arrest during G2 of the cell cycle under X-ray irradiation, DeFrank et al (1996) show that UV radiation arrests the cell cycle in G1, slowing the transit of cells into the S-phase, which is reduced by the application of caffeine. DeFrank et al. investigate the role of the p53 tumour suppressor protein whose function is inactivated in malignant cells. They find that p53-null cells are more sensitive to UV light, only in the presence of caffeine, implicating caffeine in processes which reduce cell repair and enhance cell damage under UV exposure.

The role of caffeine in accelerating mitosis before DNA repair can take place implicates caffeine in enhancing chromosome aberrations. This shows the quite complex

interactions of diet, environmental exposure and other factors such as familial genetics, in the susceptibility of people to adverse health effects.

4.7 EMR alteration of the cell cycle time:

Brulfert et al. (1985) studied the growth of plant roots in 2 day exposure to a strong (430 V/m) ELF (60 Hz) electric field *in vivo*. They found that exposed roots were shorter because cell elongation was reduced in exposed roots compared to controls. Heller and Teixeira-Pinto (1959) showed that a strong pulsed RF (27 MHz) field caused chromosome breaks which probably occurs in the replication of DNA in the S-phase. These pose the question as to whether animal cells are similarly affected.

Levin and Ernst (1995) report that 60 Hz fields (3.4-8.8 mT) and magnetic fields over the range DC-600 kHz (2.5-6.5 mT) can alter the early embryonic sea urchin embryos by inducing alterations in the timing of the cell cycle. Their results, as for the cellular studies above, were dose-dependent and biphasic as a function of frequency, duration and timing of the exposure. Low frequencies advanced mitosis and higher frequencies delayed mitosis. Stein and Lian (1992) point out the importance of cell cycle perturbations since the loss of growth control in transformed and tumour cells is accompanied by an abrogation of developmental regulatory mechanisms that are functionally coupled to proliferation.

Do the differences of these sea urchin cells and human cells mean that people will not experience alteration of their cell cycle in exposure to EMR ?

Conti et al. (1983) investigated the effects of extremely low frequency EMR on immature human peripheral blood lymphocytes which were also exposed to substances which participate in the mitosis of the cells. They found that a frequency window (3-50 Hz) significantly inhibited the conA-induced blastogenesis, while the pokeweed mitogen (PWM) was significantly affected only at 3 Hz. Conti et al. explored the mechanisms which EMR might have and excluded a direct effect on thymidine incorporation. They focus on the flux of calcium ions. A reduction of calcium ions upon exposure to EMR (through the outward flow through the cell membrane - calcium ion efflux). The effect on lymphocytes of calcium loss represents a decrease in the rate of DNA synthesis in all cells and/or a reduction in the number of cells undergoing DNA replication.

Hence they conclude: "Ca^{2+} ions are involved in the control of lymphocyte proliferation. In fact, mitogenic lectins produce a rapid, initial calcium influx and calcium is required for DNA synthesis some 18-72 h after the mitogenic stimulus." Considering the theoretical (and observed) effects of EMR on cellular efflux of calcium ions "we think that an alteration of calcium fluxes by EMF may be the most realistic hypothesis to explain the observed inhibitory effect on human lymphocyte blasticgenesis."

Human lymphocytes are the primary agents in the immune system, in the form of T-cells, B-cells and NK-cells (natural killer). The calcium ion efflux is now well documented to increase with ELF modulated RF signals and hence these signals, at levels down to SAR of 0.00015 W/kg, Schwartz et al. (1988) have the effect of reducing the protection to infection offered by the immune system cells (white blood cells), Walleczek (1992). This corresponds to an energy flux of about 0.04 $\mu W/cm^2$ for isolated frog hearts or about 0.4 $\mu W/cm^2$ for a human body.

Professor Stephen Cleary's group has been studying the effects of cell cycle changes when exposed to RF (27 MHz) and MW (2.45 GHz) radiation. Clearly et al. (1990a) exposed human blood to these EMR frequencies under isothermal conditions (37 ± 0.2°C) for 2 h and observed a statistically significant biphasic, dose-response dependent effects of the radiation on human lymphocyte proliferation, both with and without a mitogenic stimulation.

Cleary et al. (1990b) carried out a similar exposure experiment on human glioma cells. They found alterations of the cell proliferation which were not caused by RF-induced cell heating. The dose-response for both frequencies was biphasic. Lower exposures enhanced proliferation while exposures over 50 W/kg suppressed DNA and RNA synthesis. Statistically significant time-dependent alterations were detected up to 5 days postexposure, suggesting a kinetic cellular response to RF radiation and the possibility of cumulative effects of cell proliferation. Cleary et al. (1992) concluded that there is direct, nonthermal cellular effects of RF radiation which included effects on the mitotic cell cycle but no mechanisms had been identified.

Cleary et al. (1996) exposed Chinese hamster ovary (CHO) cells to within 37+/-0.1°C, to 5 W/kg and 25 W/kg signals of 27 MHz and 2.45 GHz radiation. They studied the effects at each phase of the cell cycle, including DNA distributions. They found that a 2hr exposure induced significant time-dependent cell cycle alterations for up to about 4 days. These effects were generally reversible over 96 hours and were twice as great for the 2.45 GHz microwave signal as they were for the 27 MHz radiofrequency signal. They considered this to be a real effect of relatively low magnitude and in "agreement with predictions of a theoretical analysis", referring to Liu and Cleary 1995.

4.8 Conclusion:

RF and MW electromagnetic radiation has frequency and intensity effects on the alteration of cell cycles which are not heat induced effects. A highly probable mechanism involves the change in cell membrane permeability to key agents. For example, the efflux of calcium ions which alters the synthesis of DNA and other aspects of the cell cycle in plant, animal and human cells, including cells of the central nervous system, immune system and cardiac system.

5. Cellular Control Factors

5.1 Gap-Junction Communication:

Cell-to-cell communication takes place through signals transmitted through the intercellular fluid, and through direct cell-to-cell contact through two apposed epithelial cells made of two hexagonal studs embedded in the membrane layer, called a Gap Junction.

Through this structure, ions, amino acids, sugars, nucleotides and other molecules which are smaller than 20 Å in diameter pass, but proteins, nucleic acids and larger molecules cannot, from Bretscher (1985).

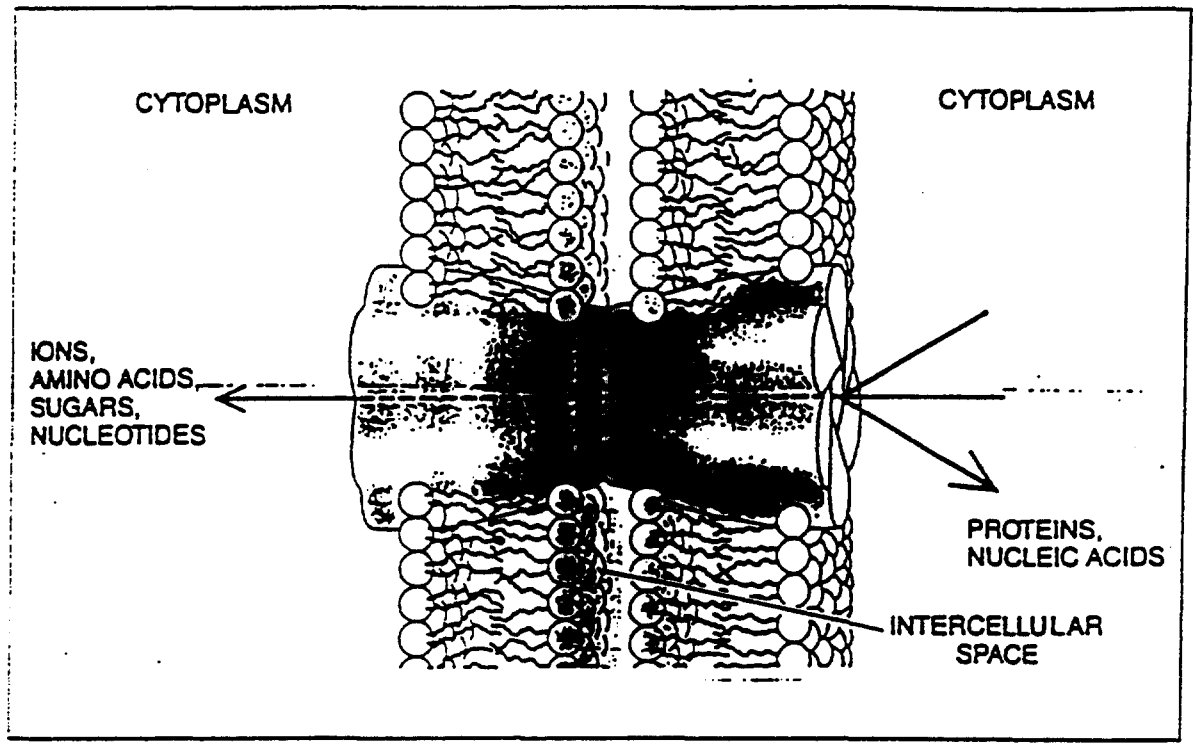

Figure 12: Gap Junction between adjacent cells (Schematic) from Bretscher (1985).

5.2 Gap Junction Alteration by EMR:

A 2 mT 50 Hz field induced a 160 % flow of cAMP through gap junctions in a monolayer of mouse fibroblast cells, measured immediately after a 5 minute exposure, Schimmelpfeng et al. (1995). Cyclic AMP is a primary "second messenger" of the cell developmental biochemistry along side calcium ions. Cooper (1995) calculated that millisecond applications of electric fields on the range 0.01 to 0.1 V/cm can result in significant hyperpolarizations and depolarizations across the gap junction. Cell-to-cell is a vital biological function. Disruption of the gap junction communication is associated with unregulated cell growth, Adey (1989).

Fletcher et al. (1987) noted that the blockage of the entry of natural cytolytic substances, alpha-lymphotoxin (LT) and recombinant tumour necrosis factor (TNF), into Chinese hamster ovary cells depends on their ability to form gap junctions, a function which varies between different strains of these cells. Fletcher found that the phorbol ester cancer promoter (TPA) opens gap-junctions to permit the entry of LT, leading to cell death (lysis) in a dose-dependent fashion.

Weak RF fields (450 MHz, 1-1.5 mW/cm^2 incident energy) with 16 Hz sinusoidal modulation, enhanced this ability of TPA to impair gap-junction communication. The effect did not occur without modulation.

Oncogenes may also interrupt gap-junction communication. Hence, EMR modifies gap-junction communication in ways which are potentially adverse to the health of tissue, either through cell death or through disrupted growth control which leads to cancer cells.

5.3 Extra-cellular environment:

In cellular aggregates that form tissues of higher animals, cells are separated by narrow fluid channels that take on a special importance in signaling from cell to cell. These channels act as windows on the electrochemical world surrounding each cell. Hormones, antibodies, neurotransmitters and chemical cancer promoters, for example, move along them to reach binding sites on cell membrane receptors, Adey (1992a). These narrow fluid "gutters", typically not more than 150 Å wide, are also preferred pathways for intrinsic and environmental electromagnetic (EM) fields since they offer a much lower electrical impedance than cell membranes. Although this intercellular space (ICS) forms only 10 % of the conducting cross section of typical tissue, it carries at least 90 % of any imposed or intrinsic current, directing it along cell membrane surfaces.

5.4 Signal Transduction:

The division of labour among the cells of a multicellular organism requires that each cell population be able to call on the services of some cell populations and respond to the requirements of others. Much of this is accomplished with chemical and electrical signals. Yet most of the arriving signals never invade the privacy of the cell. They are picked up on the surface of the cell by molecular antennae called receptors. This initiates the communication into the cell in a process termed "signal transduction".

> **"Signal Transduction refers to reactions by which the cell receives and acts upon regulatory information from outside the cell. Information-containing signals may include neural messages, hormones, growth regulatory factors, chemical substances, physical forces, and electromagnetic variables such as heat, light, and internal currents from bones and muscles. Signal Transduction is very specific and sensitive. Only particular cells respond to signals and some signal transduction systems can amplify the incoming signal by many orders of magnitude, for example a single photon of light in the eye can induce the synthesis of millions of molecules of neurotransmitters in the nerves leading from the eye to the brain (Stryer, 1986)."**

5.5 Signal Transduction structures:

Numerous stranded protein molecules protrude from within the cell into this narrow ICS. Their glycoprotein tips form the glycocalyx which senses the chemical and electrical signals in the surrounding fluid. Their highly negatively charges tips form receptor sites for hormones, antibodies, neurotransmitters and for many metabolic agents, including cancer promoters.

These charged terminals form an anatomical substrate for the first detection of weak electrochemical oscillations in the pericellular fluid, including the field potentials arising from activity of adjacent cells or as tissue components of environmental EM fields, Adey (1993).

A schematic of the cell plasma membrane is given in Figure 13, from Bretscher (1985).

These stranded protein molecules are the structures providing signal transduction of biochemical messages into the cell to alter cell metabolism or behaviour as a response to external (cell to cell, or environmentally sourced) stimuli.

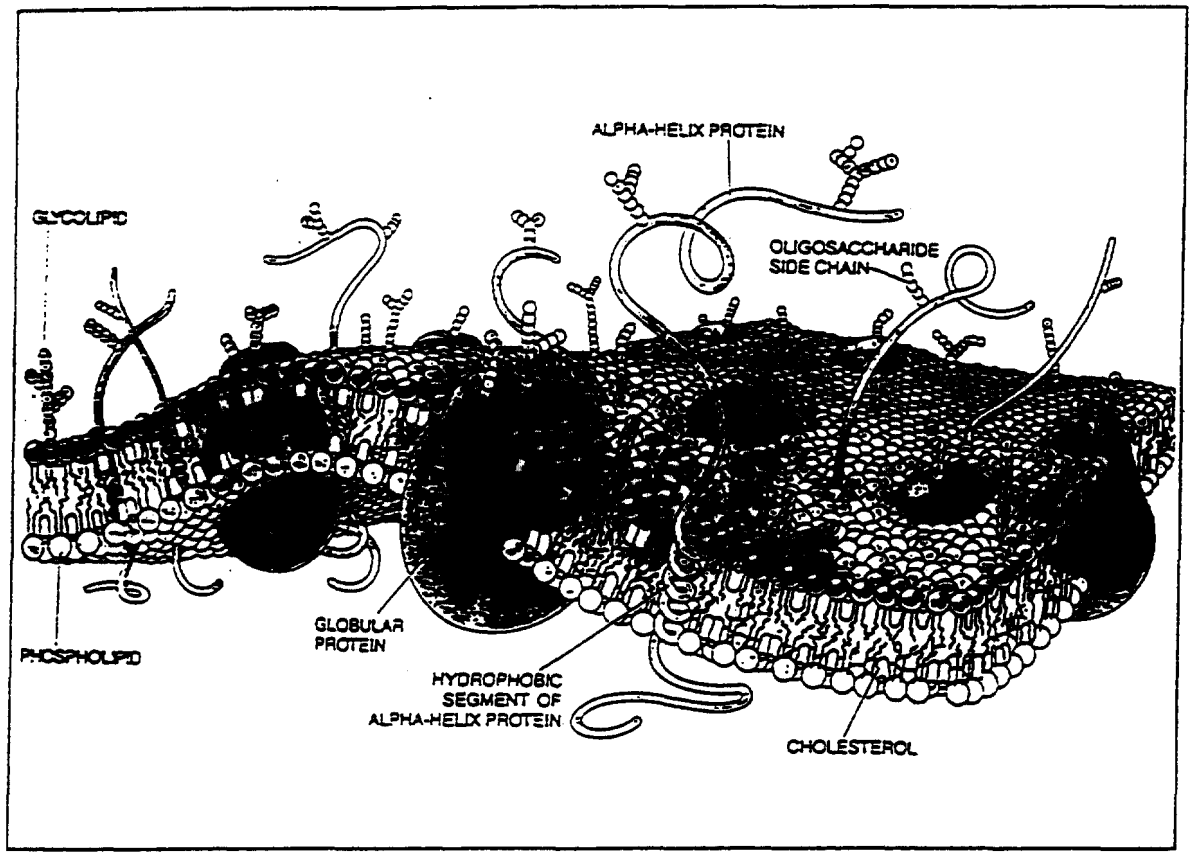

Figure 13: Cell membrane (schematic) showing the bimolecular layer in which cholesterol and other protein molecules are imbedded, with the stranded (alpha-helix) protein which has a coiled hydrophobic section within the membrane and "Y" shaped receptor sites on the extracellular strands, Bretscher (1985).

The amino acid sequence of these stranded proteins reveals a hygrophobic segment of 23 amino acids in the portion which passes through the cell membrane, and the response of these strands to epidermal growth factor (EGF) results in the proposition that this short segment produces vibration modes in the helical proteins which act as a nonlinear amplifier of the signal, Ullrich et al. (1985), Lawrence and Adey (1982).

5.6 Signal transduction messengers:

An external signal (first messenger) is provided by a messenger binding to a receptor on the stranded protein. The alpha helix transfers the message by changing shape successively down its length. At some point the signal is transferred to ions or chemicals in the cytoplasm through the action of an "amplifier" enzyme. A typical amplifier process involves adenylate cyclase which converts adenosine triphosphate (ATP) to cyclic adenosine monophosphate (AMP) (cAMP) by removing two of the three phosphate groups. ATP serves the cell by donating energy to chemical reactions. The

phosphate groups. ATP serves the cell by donating energy to chemical reactions. The intracellular signals are carried by "second messengers" such as cAMP.

The number of second messengers is surprisingly small, in other words, the intracellular signal pathways are remarkably universal. Yet the known messengers are capable of regulating a vast variety of physiological and biochemical processes.

Three of the major signal pathways are:

1. The Adenylate cyclase pathway, converting ATP to cAMP, both enhanced by stimulation and reduced by inhibition. This also modifies the calcium pathway.

2. The calcium ion, IP_3 and DG, pathway. This plays a central role in the regulation of cell growth and is not known to be inhibited.

3. The polyamine pathway, which involves the enzyme ornithine decarboxylase (ODC).

Figure 14 shows a schematic of the first two of these signal transduction pathways.

The third signal transduction pathway involves polyamine biosynthesis. The polyamines are found ubiquitously in nature and have been closely linked to the processes of cell proliferation, hypertrophy and differentiation in eukaryotic cells, Byus (1994). (Eukaryotic cells are cells of higher plant and animals, having a true nucleus). ODC decarboxylates, or removes, the carboxyl group from ornithine to yield putrecine or diainobutane, and by a further series of reaction yields spermidine and spermine, Byus (1994). Enhancement of Ornithine decarboxylase (ODC), the key regulatory enzyme in mammalian polyamine biosynthesis, is rapidly induced by mitogens and tumor promoters, Mar et al. (1995).

Elevated levels of ODC have been found in a number of animal and human tumours, for example stomach, colon and esophagus, Yoshida et al. (1992). A detailed analysis of ODC in Human Colon Cancer suggests that ODC activity is influenced by kinase activity, with protein kinase C being the most likely candidate, Sumiyoshi et al. (1991). Mustelin et al. (1987) have shown that ODC is also linked to T-cells membrane so that activation of ODC can be linked to neoplastic changes in cells and to alteration of immune system cells.

Due to the high sensitivity of this enzyme (ODC) to a large variety of stimuli and the involvement of changes in ODC activity and polyamines in a variety of pathologies, including cancer, ODC appeared to be a logical choice to investigate as a potential marker of exposure of cells or tissues to low-energy electromagnetic fields, Byus (1994).

Given the fundamental role of the signal transduction processes in the regulation and control of cell processes, including proliferation which occurs in cancer cells, and in the development of all cells in human bodies including brain, CNS and immune system, any evidence of changes in these processes because of exposure to environmental electromagnetic radiation is of grave concern.

The research reported below documents many induced changes at the cellular level due to EMR exposure. This follows with epidemiology showing increased health risks associated with increased EMR exposure.

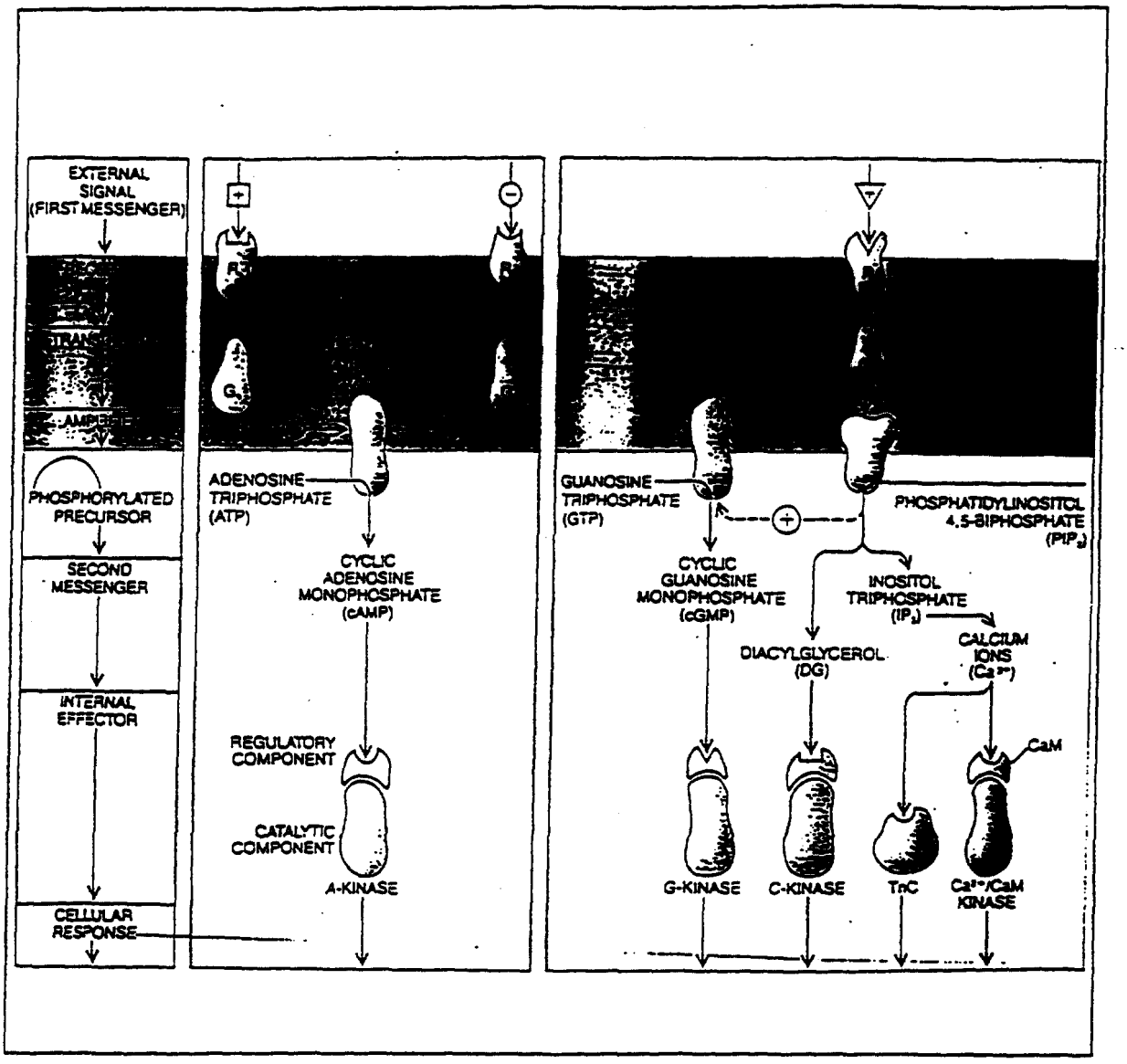

Figure 14: Schematic of two of the signal transduction pathways, with the general scheme on the left, the cAMP pathways in the centre and the DG/IP_3/Ca^{2+} pathway on the right, Bretscher (1985).

5.7 Signal Transduction Alteration by EMR:

Many laboratories have now observed increases in the enzyme ODC in cultured cells following a variety of electromagnetic fields, from pulsed static and ELF fields to modulated microwave fields. At least six separate laboratories have observed changes in ODC activity comparable to what is reported here when monolayer cultured cells were exposed to a number of ELF exposure paradigms, including pulsed electromagnetic fields, 50 Hz amplitude modulated 450 MHz fields, and 50-65 Hz electromagnetic fields, Byus (1994).

Luben (1995) summarizes the concept of signal transduction in cells and the effect of EMR:

"Clearly, any environmental influence (e.g. electromagnetic fields) that modifies signal transduction pathways in normal cells could also influence

the potentially tumorigenic pathways in susceptible cells, either by enhancing the likelihood of transformation by other tumorigenic stimuli of by acting in a direct tumorigenic manner. Thus, it is not necessary to hypothesize, as some have done, that EMF must cause genetic damage to cells in order to cause cancer or developmental abnormalities. Nor is it necessary to hypothesize that EMF must alter the expression of genes in cells directly (indeed, recent studies make this hypothesis seem rather unlikely).

By influencing signal transduction pathways, which in turn can generate cell proliferation, cell differentiation, and even transformation to a cancer phenotype, EMF can potentially be involved in a host of disease processes without ever penetrating the cell membrane in any significant manner."

There is clear biological evidence at the cellular and molecular level which shows that RF/MW radiation can be absorbed at very low levels and produce significant changes in cell behaviour and structure through signal transduction, including carcinogenic activity. Galvanovskis et al. (1996) show that modulated EMR fields change the concentrations of calcium ions and their oscillations in human leukaemia T-cells. T lymphocytes (T-cells) recognize intracellular antigens, presented at the surface of the cells. Thus there are biological mechanisms which could be related to the production of childhood leukaemia as identified through epidemiology.

5.8 Alterations in Ornithine Decarboxylase activity:

Being a frequency-related effect, the degree of coherence of the ELF signal or the modulation frequency can be relevant. Litovitz et al. (1993) investigated this matter using L929 mouse fibroblast cells exposed to 915 MHz microwaves modulated at 55, 60 and 65 Hz, with an SAR of 2.5 W/kg. They found that, as for ELF signals, a period of coherence of about 10 s was required to gain the full ODC enhancement. Litovitz et al. (1994), using a 60 Hz signal, imposed noise containing frequencies from 30 to 90 Hz.

They determined that full ODC enhancement was obtained when the rms value of the noise was less than one tenth of that of the coherent signal. These results could well have been influenced by the thermal noise of the rather intense microwave signal used. Referring back to table 2, no effect was found for calcium-ion efflux at 0.2 W/kg and higher but very significant effects were found between 0.00015 and 0.075 W/kg.

Byus et al. (1987) investigated the ODC activity in a number of established cell lines under the influence of low-energy 60 Hz EM fields. They used a 1 hr exposure to a 10 mV/cm 60 Hz field which produced a 5-fold increase in ODC activity in human lymphoma CEM cells and a 2- to 3-fold increase in mouse myeloma cells (P3) relative to unexposed cultures. Depending on the cell type, the ODC activity remain elevated for several hours after the 1 hr exposure had ceased. Reuber H35 hepatoma cells grown in monolayer culture had a 30 % increase in ODC activity with a 0.1 mV/cm field applied for 1 hr, but no effect from a 10 mV/m field applied for 2 or 3 hrs. This is another example where high intensities find no change but lower intensities do cause biological changes. Hence, while results vary with exposure interval and field strength, this shows that EMR alters ODC

intensities find no change but lower intensities do cause biological changes. Hence, while results vary with exposure interval and field strength, this shows that EMR alters ODC activity in such a way that 60 Hz fields are shown to have the potential ability as a tumour promoting stimulus in the same way that ELF modulated RF/MW also does.

Note, all the experiments reported by Byus et al. (1987) were carried out at 60 Hz and no other frequencies were investigated.

Byus et al. (1988) showed a 50 % increase in ODC activity in Reuber H35 hepatoma cells with 450 MHz microwaves modulated at 16 Hz for 1 h. This was an athermal exposure, giving less than 0.1°C temperature rise, with a 1 mW/cm^2 peak-envelope-power and an SAR of 0.08 W/kg. With σ=1.2 S/m, Eq. 11 estimates the exposure as S=35 μW/cm^2. The effect persisted for several hours following exposure. Modulation frequencies of 60 Hz and 100 Hz had no effect. A phorbol ester tumour promoter (TPA) enhanced the ODC activity in combination with the EMR. Similar ODC activity changes were observed when Chinese Hamster ovary cells and 294T melanoma cells were exposed to the radiofrequency EMR regime.

While the mechanism by which EM fields increase ODC activity is still unknown, from the observation that brief exposure of cells to EM fields altered the cell's responsiveness to TPA, and the fact that TPA has a specific receptor in the membranes of all cells, this suggests that this, and other data, are consistent with the concept that protein kinase C in the membrane may be a target for low energy EM fields.

The observation, Balcer-Kubiczek and Harrison (1985), that prior exposure to microwaves (2.45 GHz, 130 pps) led to the enhanced effect of benzpyrene- or X-ray-induced transformation frequencies, provided the cells were treated with TPA, is also consistent with the hypothesis that primary cellular effects of low level microwave fields and of TPA, is at the level of the cell membrane.

Balcer-Kubiczek and Harrison (1985) conclude that this is further evidence that microwaves are cancer promoters using mechanisms which are athermal and act at the cell membrane level. Direct application of this to animal and human cancer is found in Sumiyoshi et at. (1991). They state:

> **"ODC is a rate-limiting enzyme in the biosynthesis of polyamines linked with normal and neoplastic cell proliferation. Induction of ODC has been suggested to play an important role in tumor including skin, urinary bladder, stomach and colon carginogenisis in rodent models. ...**
>
> **Studies have shown that human colonic mucosal levels of ODC activity are lowest in colonic mucosa from healthy controls but are increased in normal-appearing mucosa from subjects with colonic polyps and from colon cancer patients."**

Yoshida et al. (1992) , investigating levels of ODC gene in human cancers, found that the ratios of ODC mRNA in tumours compared to normal tissue was 14.6 ± 3.7 for all esophageal cancers, 2.9 ± 0.9 for stomach cancer, 2.1 ± 0.9 for colon cancers, and 0.9 ± 0.2 for liver tumours.

5.9 ODC Summary and Conclusions:

Research shows that ODC, a growth regulating enzyme in the polyamine signal transduction pathway, is enhanced in a number of cell lines, including human cells, in the presence of ELF or ELF modulated RF/MW radiation. The mechanism is at yet unknown but could well involve protein kinase C in a receptor on the surface of the cell membrane.

This is relevant to the effects of signal transduction pathways on the formation and promotion of cancer for it is found that ODC levels are highly elevated in neoplastic tissue in many human cancers. The relationships between signal transduction processes, cell growth, differentiation and neoplastic transformation of cells is very complex.

What is relevant here is that many genes known to be oncogenes are clearly analogous to membrane receptors or to molecules involved in the signal transduction pathways activated by membrane receptors. Intracellular regulatory pathways such as the cell division cycle and the promotion of differentiation and gene expression are very likely to be modulated by a multitude of signal transduction pathways in both normal cells and in neoplastically transformed cells, Luben (1995).

Clearly, any environmental influence, such as electromagnetic fields, that modifies signal transduction pathways in normal cells could also influence the potentially tumorigenic pathways in susceptible cells, either by enhancing the likelihood of transformation by other tumorigenic stimuli or by acting in a direct tumorigenic manner.

Thus it is not necessary to hypothesize, as some have done, that EMF must cause genetic damage directly to cells in order to cause cancer or developmental abnormalities. Nor is it necessary to hypothesize that EMF must alter the expression of genes in cells directly. By influencing the signal transduction pathways, which in turn can regulate cell proliferation, cell differentiation and even transformation to a cancer phenotype, EMF can potentially be involved in a host of disease processes without ever penetrating the cell membrane in any significant manner, Luben (1995).

5.10 Central Nervous System Cells and EEG:

Typical electroencephalographs (EEG) are given below, from Dorland 28, p535. As noted above, cells have electric charges on their surfaces. By placing sensor electrodes on the surface of the skull electrical signals are detected. Voltages are detected which have been produces by currents emanating from nerve cells in the brain. The dominant frequency of these signals in about 8 to 10 Hz with an amplitude of 10 to 100 μV.

EEGs are characterised by frequency bands which are associated with various brain states, Dorland 28:

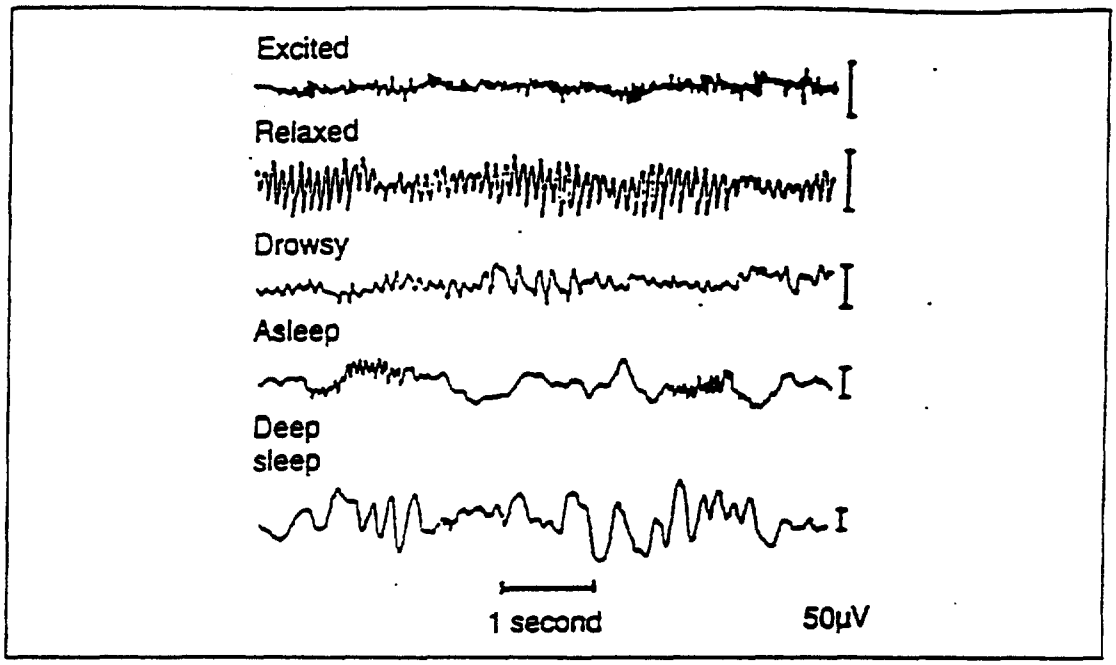

Figure 15: EEG recordings made while the subject was excited, relaxed and in various stages of sleep. During excitement the brain waves are rapid and small amplitude, whereas in sleep they are much slower and of greater amplitude.

Alpha Rhythm: 8 - 13 Hz

Are typical of the normal person awake and in a quite resting state, and principally in the occipital region. Alpha amplitude increases with joy and anger and decreases with fear and sorrow.

Beta Rhythm: 18 - 30 Hz.

Are typical during periods of intense activity of the nervous system, occurring principally in the parietal and frontal regions.

Delta Rhythm: < 3.5 Hz.

Typically occurs in deep sleep, in infancy and in serious brain disorders.

Theta Rhythm: 4 - 7 Hz.

Occurs mainly in children but also in adults during periods of emotional stress.

Cells of the brain and central nervous system which are involved in communication between neurons are dendridic. Their long dendrites house axons and receptors which release and receive messages, and from which neurotransmitters are released and to which they absorbed, Figure 16.

Adey (1979) notes that the electric process between dendrites (in the brain) is one of slow waves, not pulses.The integral of the slow wave activity of the dendrites constitutes

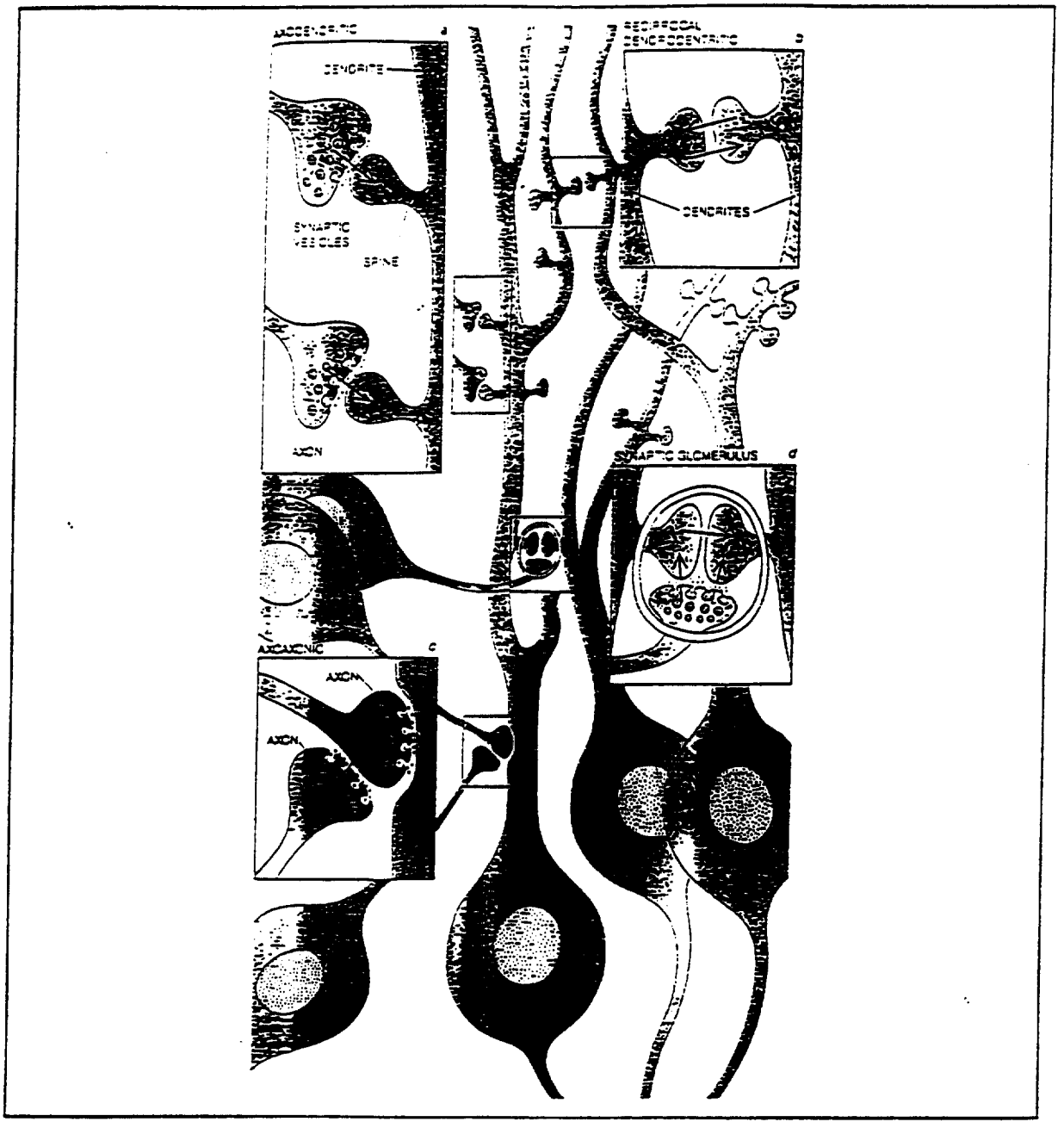

Figure 16: Communication between neurons takes place across gaps called synapses. In classical axodendritic synapse (a) synaptic vesicles in the axon of one neuron release neurotransmitter toward the receptors on the dendrite of a target neuron. It is also possible for a dendrite to pass a message to another dendrite by way of dendrodenritic synapses. In a reciprocal dendrodenritic synapses (b) each dendrite passes messages to the other by way of a separate synapse. In some synapses, called axoaxonic synapses (c), the axon of one neuron passes a message through the axon of another neuron to the dendrite of a third neuron. In synaptic glomerulus (d) the axon of one neuron passes messages to dendrites of two others; the dendrites may pass messages to each other as well. Snyder (1985) [p138]

the electroencephalogram (EEG). When the brain is awake the electro-encephalogram is

leakage of these big waves from inside the dendrites into the fluid around the cell. The electroencephalogram recorded over the dimensions of the cell is a few microvolts. The neuronal wave inside the cell is of the order of 5 to 15 mV. This the difference in amplitudes is about 200 to 1.

It is very evident that brain activity changes a great deal with rest and activity, with health and illness, and with stress and emotion. During these wide ranging changes, significant changes in neurotransmitters such as serotonin and adrenaline have also been monitored. These biochemical changes send neurohormones throughout the body to change heart beat, vasodilation etc.. They also change the brain cell behaviour in such a way that electrical signals in regions of the brain show altered, coordinated and repeatable changes in oscillating voltages which are indicative of coordinated electrical communication between large groups of brain cells.

A key scientific question is: is the EEG simply a product of the changing electrical environment within the brain, or does it provide the opportunity for external oscillating electrical fields to superimpose changes in the electrical behaviour of the brain which would then produce imposed changes in neurotransmitters and neurohormone production? Is the brain, or parts of the brain, sensors which can pick up external electrical signals which can change the psychological and/or physiological state of the brain and body ?

Professor Adey, and others, have been able to show that imposed oscillating electromagnetic fields can produce significant and repeatable changes in the behaviour of advanced mammals (cats and monkeys) in the laboratory, Adey et al. (1979). They used 450 MHz microwave signal at 0.8 mW/cm^2, modulated at 10 Hz, which produced an EEG level voltage gradient in the cat's brain of 0.1 V/cm and no detectable heating.

Wever (1974), section 3.2 above, showed changes in human subjects isolated from environmental stimuli including ELF fields, which resulted in altered circadian rhythms which were corrected by applying a 10 Hz, 2.5 V/m field, which produces about 10^{-7} V/cm in tissue. The experiment was repeated using birds, with similar results, of lengthened circadian rhythms.

> **"RF fields that are sinusoidally amplitude modulated at ELF frequencies produce a wide range of biological interactions. Induced electric gradients can be substantially higher than those produced by simple ELF electric fields, and at levels of 10-100 mV/cm, are the same range as intrinsic oscillations generated biologically, such as the electroencephalogram (EEG).", Adey (1990)**

How does the brain cells sense these EMR fields ? The cell membrane outer surface is charged and the alpha-helix glycoprotein stands outside ends are highly charged. Calcium and hydrogen ions interact with the strands and its receptors, which is the first and most sensitive transductive coupling in brain tissue.
Many studies have shown significant efflux of calcium ions from cells exposed to ELF modulated RF and ELF fields. A similar efflux has been recorded for the amino acid neurotransmitter gamma-aminobutyric acid (GABA), Kolomytkin et al. (1994), in association with microwaves modulated at 16 Hz.

This is very significant since GABA and glutamateric synapses make up about 60 % of the CNS and calcium ions appear to hold the key to every aspect of cell-surface transduction, Adey (1979). Kolomytkin et al. (1994) showed that at 915 MHz microwave signal, modulated at 16 Hz, altered the binding of 3H-glutamate and 3H-muscimol in rats brains, at power densities below 50 $\mu W/cm^2$, which are statistically significantly different from controls to below 10 $\mu W/cm^2$, Figure 17.

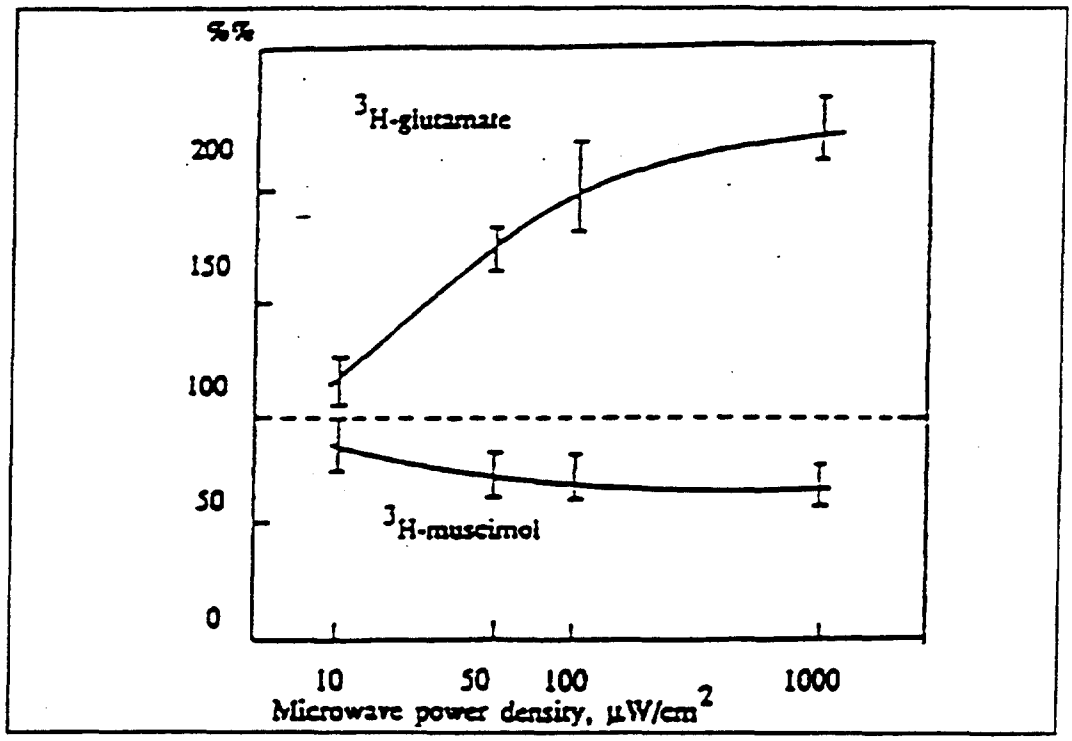

Figure 17: Altered the binding of 3H-glutamate and 3H-muscimol in rats brains versus microwave power density (915 MHz, modulated at 16 Hz), Kolomytkin et al. (1994).

Kolomytkin et al. (1994) link these changes to Ca^{2+} ions which have been shown to stimulate specific glutamate binding to synaptic membranes due to the activation of a calcium-dependent protease and resulting proteolysis (splitting into fragments) of cytoskeletal proteins.

Dumanskiy and Shandala (1974) and their colleagues reported altered conditioned reflex in rabbits and rats chronically exposed to extremely low levels of VHF and microwave fields. They used either 50 MHz or 2.5 GHz CW fields or 10 GHz 1 μs pulses at 1,000 or 20 Hz, with 10-12h daily exposure with 50 MHz and 8 h with microwave fields. They found statistically significant effects with field intensities between 1.9 and 2.0 $\mu W/cm^2$.

In each experiment the animals were irradiated for 120 days, with a 60 day follow-up. For the first 10 days the animals were "somewhat excited" and reacted to the onset of exposure. Thereafter responses to conditioned stimuli has a longer latency, with weaker responses to positive stimuli and more numerous missed responses, leading to "pathologic stagnation and inertia".

6. Calcium Ions:

6.1 Calcium Ion Processes:

6.1.1 Ionic calcium is ubiquitous in mammalian cells.

Calcium is nearly ubiquitous in human cells. Calcium ions (Ca^{2+}) play vital rolls in many biological processes of living tissues, including signal transduction processes at the cell level, which includes processes which control the binding and release of molecules to the surfaces of cells which influence primary cellular behaviour. The intracellular fluid (fluid inside the cell membrane surrounding the cellular nucleus), is rich in calcium ions. When calcium ions flow outwards through the cell membrane, it is called "calcium-ion efflux".

A molecular analysis of the cAMP pathway shows that cAMP often activates the calcium ion pathway and modulates its activity. The heart provides a now classic example. There epinephrine acts through the cyclic AMP pathway to modulate the level of intracellular calcium. This the force of each heart beat which is governed by a brief calcium pulse.

In certain cells, such as neurons, the source of calcium ions is well known: it is the extracellular fluid. Nerve signals arriving at the synaptic terminals of a neuron decrease the voltage across the neuronal cell membrane; the resulting "depolarization" opens voltage-sensitive calcium channels through the cell membrane. Before depolarization, the Ca^{2+} concentration in the cytoplasm is about 6×10^{14} ions/cc. The Ca^{2+} concentration outside the neuron is about 10,000 times higher. Hence the depolarization enables calcium ions to flood into the neuron and trigger a cell response. Even a rather small change in intracellular calcium can exert profound changes in cellular activity, Bretscher (1985). In the synaptic terminals of neurons, for example, calcium induces the release of neurotransmitter molecules.

The extracellular fluid cannot be the sole source of calcium ions. For one thing the absence of extracellular calcium does not prevent external messenger acetylcholine from stimulating the pancreas to release the digestive enzyme amylase. Thus it has become apparent that calcium employed by a cell for internal signaling not only enters the cell from outside but is also released from internal reservoirs. There turn out to be many examples of hormones or neurotransmitters employing internal calcium to control physiological processes, Bretscher (1985).

Hence external stimuli which can cause influx or efflux of calcium ions from the cell have clear and important consequences for cell growth regulation, cell death, neurotransmitter and hormone balance.

6.2 Calcium ions and Electromagnetic Interactions:

A perspective on the EM properties involved can be seen by noting that the characteristic membrane potential of most cells is about 0.1 V in a resting state. Since this exists across the very thin (40 Å) plasma membrane, it creates an enormous barrier of the order of 10^5 V/cm. However imposed ELF and amplitude modulated RF fields produce tissue gradients in the range 10^{-7} to 10^{-1} V/cm, which are gradients involved in essential physiological functions in marine vertebrates, birds and mammals, Adey

(1981). In vitro studies have reported similar sensitivities for cerebral Ca^{2+} efflux, and in a wide range of calcium-dependent processes that involve cell membrane functions, including bone growth, modulation of intercellular communication mechanisms that regulate cell growth, reduction of cell-mediated cytolytic immune responses, and modulation of intracellular enzymes in signal transduction. These processes have been confirmed for many human cell types, including lymphocytes, ovary cells, bone cells, fibroblasts, cartilage cells and nerve cells, Adey (1992a).

Since the electric field strength varies as the square root of the exposure (Eq. 3), for a 147 MHz modulated RF field with an environmental exposure of $1\mu W/cm^2$ or $0.1\mu W/cm^2$ the Tissue Gradients are estimated at $3.5x10^{-3}$ V/cm and $1.1x10^{-3}$ V/cm , respectively. These are still at least 10,000 times higher than the lower limit of 10^{-7} V/cm.

Calcium ion efflux from within cells clearly alters the intracellular calcium ion concentration, which alters the Calcium ion signal transduction process which is vital to balanced regulation of cell growth and, in neuron tissue, neurotransmitter and neurohormone production and reception. In other tissue it alters the reaction to the stimulation of antibodies because the role of calcium ion homeostasis in activation of channels of cells in the immune system.

Luben (1995) summarizes research through which RF radiation which is modulated at ELF frequencies changes the calcium ion efflux in Table 2.

It is now widely accepted that calcium plays a central role in the development of the immune response, Grinstein and Klip (1989). Changes in the cytoplasmic free calcium concentration (Ca^{2+}) are thought to be essential for responses as varied as bacterial killing by neutrophils and the synthesis and secretion of antibodies by lymphoid cells.

It is pertinent to note that the "no effects" studies of Merritt et al. (1982) are consistent with the power intensity windows identified by Blackman et al. (1980a, 1988). It is well established that calcium ion efflux changes are not linearly related to intensity, but rather to particular combinations of intensity, modulation frequency and temperature range. It is also pertinent to note that although a great deal of calcium ion efflux research has focused on ELF exposures, the table 2 above is for modulated RF/MW exposure, with effects being found for carriers in the range 50 MHz to 915 MHz and modulation frequencies in the range 0.5 to 32 Hz.

Calcium ion signaling is a function of the central nervous system (CNS). Walleczek (1992) proposes that research findings show that membrane-mediated calcium ion signaling processes are involved in the mediation of ELF effects on the immune system. ELF modulated microwaves have similar effects.

Shandala et al. (1979) found that calcium ion efflux varies in living animal cells at $10\mu W/cm^2$ using microwaves (about 0.0075 W/kg), consistent with Kolomytkin et al. (1994).

The understanding of the role of intercellular calcium ions has been growing and evolving rapidly over recent years. The fact that ELF radiation, and RF/MW radiation which is modulated at ELF frequencies, significantly alters the calcium ion concentrations and efflux in intracellular fluid is well proven and documented down to SARs of 0.00015 W/kg.